

Xenotorodor stygioxanthus gen. nov., sp. nov. (Lepidoptera, Lecithoceridae, Torodorinae), described from an established population in Spain with discussion of taxonomic placement

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Abstract. The family Lecithoceridae is not well represented in the Palaearctic region, with very few taxa in Europe. Here we describe a new genus and species of lecithocerid moth, *Xenotorodor stygioxanthus* Sterling, Lees & Grundy, **gen. nov., sp. nov.** The taxon represents a subfamily new to Europe. We consider placement of the genus within Crocanthinae or Torodorinae. We place it in Torodorinae, notwithstanding the reduced gnathos in the male genitalia. DNA barcodes suggest that the taxon belongs within a distal clade of this subfamily. They are over 9.2% pairwise divergent from any hitherto cleanly sequenced Lepidoptera taxon and over 10.1% from nearest taxonomically identified neighbours in Neighbor Joining and ML trees. Characteristics of the DNA barcode and morphology of this new taxon suggest that refinement of synapomorphies for the family and two subfamilies is needed. We have nearly 100 records for this new species since 2020, all from a small area of Southern Spain, close to the Straits of Gibraltar. The life history and early stages of the species are unknown.

Introduction

The family Lecithoceridae is a poorly known group of microlepidoptera which are found principally in the Oriental tropics, and the Australian and Afrotropical regions. Only 64 species (4.5% of the total number) are known from the Palaearctic region (Park et al. 2022) and, of these, only 10 species are reported from Europe (for these purposes the European Economic Area and other Western European countries) (<https://lepiforum.org/wiki/taxonomy/Gelechioidea/Lecithoceridae?view=0®ions=eu>). Five of these species have previously been recorded from Spain. All these European taxa belong to the Ceuthomadarinae and Lecithocerinae. Here we describe a micro moth which is genetically divergent from all hitherto DNA barcoded Lepidoptera and differs morphologically from described species of Lecithoceridae.

The taxon was discovered by Dave Grundy (DG), who found the first specimen at mercury vapour light at the research facility at the Centro Internacional de Migración de Aves (CIMA), Tarifa, Cádiz Province, Spain on 18 May 2020. Since this discovery, DG has recorded a total of 93 adult specimens from various locations within or near the research centre at CIMA and a further two adult specimens from Huerta Grande, Pelayo, Cádiz Province, also near to the coast on the

Spanish side of the Straits of Gibraltar, approximately 10 kilometres from CIMA. These have all been found at mercury vapour light or LEDs.

In seeking to identify these specimens we first considered the European taxa of Lecithoceridae (comprising two Ceuthomadarinae in the genus *Ceuthomadarus* Mann, 1864 and eight Lecithocerinae in the genera *Eurodachtha* Gozmány, 1978, *Lecithocera* Herrich-Schäffer, 1853, and *Homaloxestis* Meyrick, 1910, but here not including two Oditinae (Peleopodidae): Karsholt and Razowski 1996; Gozmány 2012; Barton 2015). However, these taxa were easily ruled out on morphological grounds. A search of images on Google revealed striking similarities in general habitus and even resting posture of the new taxon to the genera *Sisyrodon* Meyrick, 1922 and *Protolychnis* Meyrick, 1925 especially as regards the thickened antennae and wing pattern. The arrival of Park *et al.*'s (2022) global review of Torodorinae, together with Park's previous comprehensive treatment of Crocanthinae (Park 2015), allowed us to check from a morphological perspective all possible generic affiliations for those subfamilies. To go further, we DNA barcoded three specimens and subsequently analysed all existing DNA sequences of Lecithoceridae in the public domain together with the COI data for the new taxon.

On the basis of the molecular evidence set out here and comparative morphology, using a process of elimination benefitting from the comprehensive accounts of Park (2015) and Park *et al.* (2022), as well as comprehensive searches of the Gelechioidea collection at the Natural History Museum, London (NHMUK), we conclude through careful consideration of taxonomic placement that the taxon described here represents a previously unknown European lineage in the Torodorinae and a new genus.

Materials and methods

The examined specimens for this paper were obtained live from light traps, refrigerated overnight and set on the following morning. The morphology of 16 specimens of this taxon collected by Mark Sterling (MS) and DG was examined. The illustrated material was photographed using a Canon EOS 5DSR camera and MP-E 65 mm lens equipped with a Stackshot system operated by Helicon Remote software (version 3.8.4 W); the shots were eventually stacked with Helicon Focus software (version 6.7.1), which was set up with montage controlled by Helicon using a motorised deck in about 30 to 40 steps for adults and 10 to 15 steps for genitalia and wing preparations. Genitalia dissection and mounting followed Robinson (1976). Descriptions of the genitalia follow Klots (1970) and Kristensen (2003).

The DNA from three male specimens obtained from Tarifa, Cádiz, Prov. Andalucía, Spain, NHMUK013698467–9 (details in Type Material) was extracted at NHMUK from single hindlegs, and following purification of the resulting genomic DNA, standard Sanger PCR was used to amplify COI-5P and the amplicons checked visually using a gel using the same methodology as in Sterling *et al.* (2022), see also Cuber *et al.* (2023), with a mix of the Folmer primers (HCO2198, LCO1490; Folmer *et al.* 1994) and Hebert primers (Lep-F1, Lep-R1). The following steps employed third generation sequencing technology (see Cuber *et al.* 2023 for precise procedures). As part of two 96 well plates for a range of samples, a library was prepared by ligating standard Illumina indexes (unique 20 bp tags cross-referencing sample/well to up to 658 bp COI-5P fragments) to 20 bp M13 reverse tails attaching to each DNA fragment, using the Oxford Nanopore Technologies (ONT) SQR-LSK110 ligation kit. The sample fragments were then pooled and pipetted on the loading well of a single-use 200-pore ONT Flongle flow cell (R9.4.1, FLO-FLG001) that had previously been primed by hand pipetting of the supplied buffer. The Flongle was then fitted to a GRIDION X5 benchtop sequencing machine (<https://nanoporetech.com/products/gridion>) and run for 72 hours. A single strand was read

through by the machine singly base-by-base using ion current disturbance technology. ONT barcode software (ONTbarcode v0.1.9: <https://github.com/asrivathsan/ONTbarcoder>; Srivathsan et al. 2021) was then used in the bioinformatic pipeline for retrieving the tagged DNA barcode fragments for each sample. Between 271–388 sequences were used during this demultiplexing process, to achieve a minimum coverage of N25 (25 fragments per DNA barcode at high fidelity for consensus base calls) for each 658 bp sequence. Sequences are available in the public project DS-LECITH (http://v4.boldsystems.org/index.php/MAS_Management_DataConsole?codes=DS-LECITH) with Process Ids UKMOT004-23, UKMOT005-23, UKMOT006-23 and BIN BOLD:AFA0579 on BOLD and Accession numbers OQ339151, OQ339152, OQ339153 respectively, on GenBank.

We first checked the global database of DNA barcodes on BOLD using the Identification Engine (https://v4.boldsystems.org/index.php/IDS_OpenIdEngine) and building the corresponding Neighbor Joining tree. This led to a more detailed molecular examination of the information content in DNA barcodes and tree building analyses using COI-5P.

We downloaded available Lecithoceridae from BOLD on 21/12/2022 for comparative analysis with the DNA barcode of the query taxon. We downloaded from GenBank the mixed COI and seven-gene nuclear datasets for Lecithoceridae of Kaila et al. (2011) and Wang and Li (2020) as well as three sequences for *Homaloxestis croceata* Gozmány, 1978 from the study of Regier et al. (2013) along with their DNA barcodes from BOLD. Alignment was done gene by gene using MAFFT online (Q-Ins-I option; <https://mafft.cbrc.jp/alignment/server>). We added all 17 Lecithoceridae exemplars on GenBank from these datasets (here excluding the highly divergent genus *Martyringa* Busck, 1902, as more closely related sequences, less susceptible to long branch attraction, could be used for rooting). Sequences were concatenated for the aligned genes against their sample numbers in MS Excel.

For the Lecithoceridae DNA barcodes, 435 unique BIN (Barcode Index Number) representatives that each had the longest sequence length in the corresponding ‘tsv’ file that had been downloaded from BOLD along with their GenBank accession numbers, where available, were considered and a few obvious non-members of Lecithoceridae were eliminated. We carried out a similar process for the related families Autostichidae and Xyloryctidae. Pairwise divergences were computed in this program using the Pairwise Alignment option in Bioedit 7.2.5 (Hall 1999) (‘Calculate Identity/Similarity for two sequences’) for comparable nucleotides/codons only, whereas codons were analysed using the ‘Conservation Plot’ option to a reference sequence that was edited to show the triplet ‘NNN’ for potentially informative nucleotides (reading as X for codons), and their frequencies calculated using copy/paste into a column in MS Excel. We directly examined the DNA barcodes using Bioedit alongside the DNA barcodes of the query taxon to see if there were characters linking it to particular groupings, or any character that distinguished Lecithoceridae in general. Considering these 435 BINs, six could be eliminated as obvious representatives of other families: *Rhizosthenes falciformis* Meyrick, 1935 (BOLD:AAX8698), see Wang and Li (2020: 8); the BINs BOLD:ACU2376 and BOLD:ADH8338 represent other Peleopodidae: Oditinae; BOLD:AAH3806 represents a Cosmopterigidae: Scaeosophinae; BOLD:AEA2583 represents a Cemiostomidae and BOLD:AAJ5084, NSWHP3227-19 represents a Stathmopodidae (not a *Crocantthes* Meyrick, 1886), leaving 429 BINs for further analysis. For the family level analysis we similarly considered 221 BINs in Autostichidae and 810 BINs representing Xyloryctidae.

A local BLAST was conducted in Bioedit of the (up to 658 bp) dataset constructed from these 429 sequences, to find the nearest hits, and the pairwise divergences were checked for these over comparable codons as described above.

To go further, we examined the placement of our sequences in relation to the GenBank and BOLD datasets. The idea was, when adding available DNA barcodes of Lecithoceridae, to provide a provisional scaffold for the families and subfamilies, whilst avoiding problems of paralogy by having a COI part of the dataset across all taxa, analyses that we detail here.

For the nuclear data we eliminated IDH as having only three representatives of only two subfamilies, but no Torodorinae. We analysed the resulting 5408 bp alignment (COI 1475 bp including COI-5P for 12 representatives, EF-1 α 985 bp for 15, Wingless 400 bp for 16, RpS5 600 bp for 12, CAD 850 bp for nine, MDH 407 bp for 10, and GAPDH 691 bp for six representatives). For a quick tree analysis we used Phyml 3.0 (online: <http://www.atgc-montpellier.fr/phyml>), with the GTR + G + I model selection as implemented by the Bayesian Information Criterion (BIC), and showing ABayes support.

Phyml runs were done by building datasets of publicly available DNA barcode sequences from BOLD that had been identified or were identifiable to at least generic level, alongside the 5408 bp alignment for 17 taxa, selecting where possible the longest public domain sequence within one representative of each Barcode Index Number (BIN), along with the mixed seven-gene COI and nuclear alignment. We concentrated on being as comprehensive as possible within these constraints for Torodorinae and Crocanthinae (testing between 21 and 32 terminals), ending up with a 51-terminal dataset.

For further analyses of the 5408 bp Lecithoceridae alignment from GenBank, we used IQ-TREE (<http://www.iqtree.org>) on the 17-taxon portion of the dataset, with or without the two haplotypes of the DNA barcode of the query taxon, using a partitioning file specifying the start and ends of each of the seven genes and using automatic selection (Bayesian Information Criterion) to allocate the optimal model for each gene, displaying both ABayes and Bootstrap values (100 runs), with parameters edge-linked or edge-unlinked across the partition.

Finally, we carried out a gene-partitioned analysis of the entire dataset (51 taxa, 5408 characters) in MrBayes 3.1. This analysis implements the GTR model as in Phyml but allows rates to vary among partitions using a rate multiplier. We used the following parameter settings: nst=6 and rates=invgamma. In one run we specified unlinking of the parameters statefreq, revmat, shape, and pinvar, whereas in the second, they remained linked (by default). In both cases, a variable rate prior was set, the run included 10,000,000 generations under a Markov Chain Monte Carlo process, and the burnin rejected the first 25% of trees in four chains, finally verifying for convergence between two tree runs. The resulting consensus trees provide a more rigorous and direct assessment of Bayesian posterior support than is estimated using ABayes in Phyml.

Suppl. material 1 provides details of all the sequences used in Fig. 1, and where they were published for the first time, if not in this paper. The underlying alignment is also available in an online repository (<https://doi.org/10.6084/m9.figshare.22242250>).

Molecular analysis results

DNA barcode query and distances, Neighbor Joining, BLAST

Query of the DNA barcodes of the query taxon (the barcode of NHMUK013698469 is identical to that of NHMUK013698468 and differs by a C as opposed to a T in position 118 of the holotype, NHMUK013698467) on BOLD placed the taxon as more than 9.06–9.22% by p-distance from any other micromoth. However, as an exception, a single gelechiid came up as the top hit (*Ephysteris diminutella* (Zeller, 1847), Process ID LON7008-18, GenBank accession MN805721, which is

8.99% by p-distance). Of its 618 bp, 62 are ambiguously coded. When these are ignored, it is 9.0% pairwise divergent (Bioedit), whereas a clean (658 bp) sequence adjacent by its Process ID code for the same species, LON7007-18 (MN805536) is 13.07% divergent to the query taxon. This sequence should ideally be removed from consideration by BOLD. All the other ten top hits were unidentified Lepidoptera (probably lecithocerids) or identified as Lecithoceridae. The nearest identified lecithocerid species at 9.2–9.38% p-distance for two different specimens was identified as ‘*Thubana exaema*’ (i.e., *Thubana exoema* (Meyrick, 1911)) from Sri Lanka (see Discussion). The two haplotypes of the query taxon were 9.55–9.71% to the nearest two sequences identified only as a lecithocerid, and 9.68–9.82% to the nearest ones identified as a *Torodora* (BOLD:AAH3804 from Australia and BOLD:ABY1674 from Vietnam). In the corresponding NJ tree, the query taxon linked, albeit with long branches, to eight nearest terminals representing five BINs all from SE Asia, two of which were identified as Lecithoceridae and one of which as *Torodora* Meyrick, 1894, whereas it fell relatively remotely from *T. exoema* among the top 99 hits.

As another distance approach, using a local BLAST in Bioedit of the 429 Lecithoceridae sequences representing different BINs downloaded from BOLD, the sequences of the query taxon were 9.57–9.73% pairwise divergent to a species of Lecithoceridae (BOLD:ACT7825, LNAUT3910-15, Malaysia), 9.8–9.95% to another Lecithoceridae (BOLD:ADV1376, GMPBS211-18, Pakistan), 10.18–10.33% to *Halolaguna subluxata* Gozmány, 1978 (BOLD:ABA2899, KF523781) and 10.1–10.35% to *Torodora aenoptera* Gozmány, 1978 (BOLD:AEG4946, MN852952), these representing the four top “hits” by local BLAST.

As a third approach, the top hit using nucleotide BLAST on Genbank on 29/01/2023 was *Halolaguna subluxata* (KF523779.1) at 9.86–9.71% divergence (depending on the haplotype of the query taxon). However, equally divergent was a member of Xyloryctidae, *Xylorycta cirrhophragma* Meyrick, 1921 (KF404885.1), with *H. subluxata* (KF523781.1) at 10.02%, showing weak signal to noise for this approach, which does not benefit from the DNA barcode dataset comprehensiveness of BOLD.

Inspection of sequences for synapomorphies

We then examined the amino acid translation of the DNA barcode region of the alignment, first examining the 429 BINs attributed to Lecithoceridae. The 161st complete codon has the state Asparagine (N), as opposed to Serine (S), which is more usual in Gelechioidea. This is the state in NHMUK013698467–9 and is particularly prevalent in Lecithoceridae. Among the Lecithoceridae BINs, the following states (using standard amino acid abbreviations) are represented: N = 91.8%, S = 7%, K = 0.9% and D = 0.2%. By contrast, among the 221 Autostichidae BINs the proportions of states were N = 0.9%, S = 94.6%, others = 0.45% (F, L, M, T, Y), whereas among the 810 BINs of Xyloryctidae, N = 2.5%, S = 71.7%, F = 9.5% and other states 16.3% (Y, M, A, T, Q, L, K, I). An ‘N’ (generally the triplet AAT although sometime AAC) was a feature of all the sequences identified to at least a Torodorinae genus, except that an apparent reversal (see Fig. 1b) to S (the triplet AGT) occurred locally in two of four *Sisyrodonta* BINs (BOLD:AAN9227, BOLD:AAO6600), and among those that belong to Lecithocerinae, a reversal to S was seen in the *Lecithocera nigrana* (Duponchel, 1835) complex (comprising one BIN and six closely related BINs from Greece, Madagascar, and Pakistan), and in two BINs of *Sarisophora* Meyrick, 1904 from Australia. We found no such clear synapomorphies at subfamily level for single nucleotide or codon positions, although some group-specific patterns in complete codon 94.

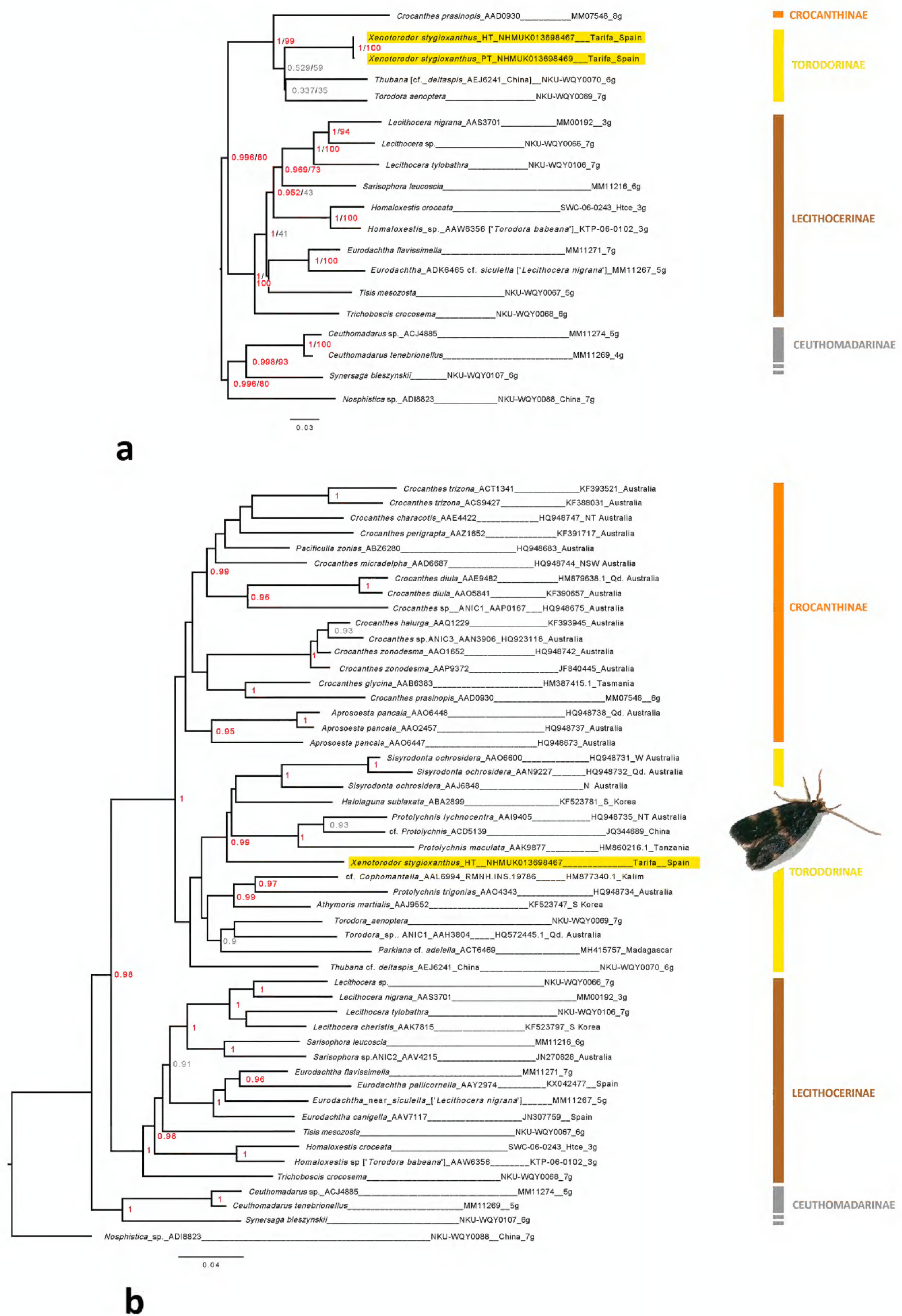


Figure 1. a. Phylogenetic analysis using ML as implemented in IQ-TREE (edge-unlinked), of Lecithoceridae using 17 taxa from GenBank for a matrix of 5408 characters and 3–8 nuclear genes ('3g'..'8g'), together with DNA barcodes of the holotype and a paratype of *Xenotorodor stygioxanthus* gen. nov., sp. nov., showing a supported position in (Crocanthinae + Torodorinae), and a branch with greater proximity to species of *Thubana* and *Torodora* than to the type species of *Crocantthes*, *C. prasinopis*. The first support value for each node is ABayes, and the second percentage of 100 bootstraps. The tree is displayed as 'midpoint' rooted.

b. Phylogenetic analysis of Lecithoceridae in Phym 3.0 showing the placement of the holotype DNA barcode of *Xenotorodor stygioxanthus* gen. nov., sp. nov. (an individual is shown in its 'alert' resting posture). The tree is based on a matrix of 51 taxa and 5408 characters, with all taxa represented by COI-5P. Terminal names include the last seven characters of the BOLD cluster (BIN, abbreviated from the format 'BOLD:ABC1234'), and the country of origin, where known or relevant. '3g'..'8g' specifies the number of genes used (two sections of COI and EF-1a are treated as single genes), for requisite taxa whose nuclear data is from GenBank. ABayes support values are shown to the right of nodes; values < 0.95 are not considered supported for the purposes of this analysis. The tree is rooted on *Nosphistica*, and all subfamilies are delineated, where known.

IQ-TREE analysis

Based on analysis of 17 terminals including nuclear data or 19 terminals including the two haplotypes of the query taxon, IQ-TREE automatically implemented the following partitioned model selection: COI: GTR+F+I+G4; EF-1a: Tim3e + G4; Wingless: K2P + G4; RpS5: TIM2e + G4; CAD: TIM2 + F + G4; GAPDH: TIM2u + F + I; MDH: TIM2e + G4. Although the edge-unlinked partitioned analysis in IQ-TREE provided a slightly higher log likelihood than the corresponding edge-linked analysis (-23644 to -23770; 17 terminals and -23805 +/-343 to -23939 +/- 346; 19 terminals) the standard errors overlapped and the lower Bayesian Information Criterion for the edge-linked analysis was therefore preferred (48548 vs 49261). In this analysis, when 17 taxa were run (i.e., only taxa with nuclear data), the Ceuthomadarinae node showed a value of pp = 1 /bootstrap = 100%, with the Lecithocerinae + Torodorinae + Crocanthinae node with 1/64%, the Crocanthinae + Torodorinae node with 1/100%, and the Torodorinae node (*Thubana* + *Torodora*) not achieving support thresholds (0.877/64%). When the two haplotypes of the query taxon were added (Fig. 1a) and 19 terminals were run, the respective nodal values became 1/100% (Ceuthomadarinae), 0.998/93% (*Ceuthomadarus* + *Synersaga*), 0.996/80% (latter including also *Nosphistica*), 1/100% (Lecithocerinae), 0.996/80% (Lecithocerinae + Crocanthinae + Torodorinae), 1/99% (Crocanthinae + Torodorinae), but unsupported (0.337/35%) for the existing Torodorinae, and no support either (0.529/59%) for a sister relationship between the new taxon and *Thubana* cf. *deltaspis* Meyrick, 1935 (Fig. 1a), although the branch of the DNA barcode of the query taxon fell closest to the torodorine rather than crocantine branch.

Phym analysis of full dataset

We attempted various phylogenetic analyses of available sequences using Phym 3.0, an instance of which is shown (Fig. 1b). We based the last analysis on a matrix of 51 taxa and the 5408 nucleotide positions, including the 17 terminals from GenBank. This dataset is contiguous for all taxa across 483 positions of COI-5P (with all but 10 exemplars contiguous over 658 bp), using the GTR + G + I model that was selected by BIC. For these 17 taxa, dispersed across the subfamilies, COI-3P and 3–7 nuclear genes were added from the GenBank dataset referred to in Materials and Methods in order to provide a general framework for rooting and for the subfamilies (number of genes shown in Fig. 1b, otherwise just COI-5P).

The Phyml analyses never placed the new taxon within the Ceuthomadarinae nor the Lecithocerinae. These groups each appear to be monophyletic according to Wang and Li 2020 and our analysis (see Park *et al.* 2022 regarding the relationship of “*Torodora babeana*” to *Homaloxestis* Meyrick, 1910). Rather, they consistently placed the DNA barcode of the new taxon within a distal group of Lecithoceridae, i.e., the subfamilies Torodorinae + Crocanthinae (in the last case comprising representatives of the genera *Crocantthes*, *Aprosoesta* Turner, 1919 and *Pacificulla* Park, 2013). Consistently the DNA barcode of the query taxon fell in a group or clade (pp = 0.99 in Fig. 1b with *Sisyrodon* ‘*ochrosidera*’—a complex of BINs, three BINs within *Protolychnis*, and *Halolaguna subluxata*. BOLD places *Sisyrodon* in Lecithocerinae, while Common 1990 has it in the Lecithoceridae, but our analyses always placed it in this grouping of Torodorinae. However, our analyses always failed to provide a significant support level for a sister genus or the sister group of the branch of the query taxon.

The Phyml 3.0 analysis only showed separation of Crocanthinae and Torodorinae in cases when taxon/BIN sampling was relatively dense (instance shown in Fig. 1b). More often with fewer terminals (especially among *Crocantthes*, which include several multi-BIN species on BOLD), resulting trees had Crocanthinae and Torodorinae partially intermingled, occasionally with the query taxon grouping with *C. diula* and *C. prasinopsis*. When the sampling of identified taxa from BOLD among Torodorinae plus Crocanthinae was improved, Torodorinae formed a separate grouping as did Crocanthinae, albeit without support. The topology of Lecithocerinae was similar to that shown by Wang and Li (2020: 5, S2, S3) except for the position of *Homaloxestis*. The query taxon fell in a supported grouping (p = 0.99) with the three *Protolychnis*, three *Sisyrodon* and *Halolaguna subluxata*.

MrBayes analysis of full dataset

The MrBayes analyses on the 51-terminal 5408 bp dataset using a paratype sequence of the query taxon (NHMUK013698469, OQ339153) provided a 50 percent majority rule consensus tree (shown in Suppl. material 2, unlinked and Suppl. material 3, linked). These analyses show support for Ceuthomadarinae (pp = 1), but for *Ceuthomadarus* + *Synersaga* only in S3 (pp = 0.96). Also, they show support for Lecithocerinae (pp = 0.99 and 1 respectively), but a lack of resolution for Torodorinae + Crocanthinae. Within this “bush”, there were a number of unsupported groupings that include the query taxon (+ *Halolaguna* + three each of *Sisyrodon* and *Protolychnis* with pp = 0.97, Suppl. material 3 or without support, Suppl. material 2), (*Athymoris* Meyrick, 1935 + *Cophomantella* Fletcher, 1940 + *Protolychnis trigonias*, pp = 0.95, Suppl. material 3 or without support, Suppl. material 2), in each case with unsupported groupings of *Crocantthes* and *Aprosoesta* (‘*pancala*’).

Morphological systematics

Xenotorodor Sterling, Lees & Grundy, gen. nov.

<https://zoobank.org/75DA3064-908C-4764-B2EC-1E4AD58C887C>

Type species. *Xenotorodor stygiexanthus* Sterling, Lees & Grundy, sp. nov.

***Xenotorodor stygioxanthus* Sterling, Lees & Grundy, sp. nov.**

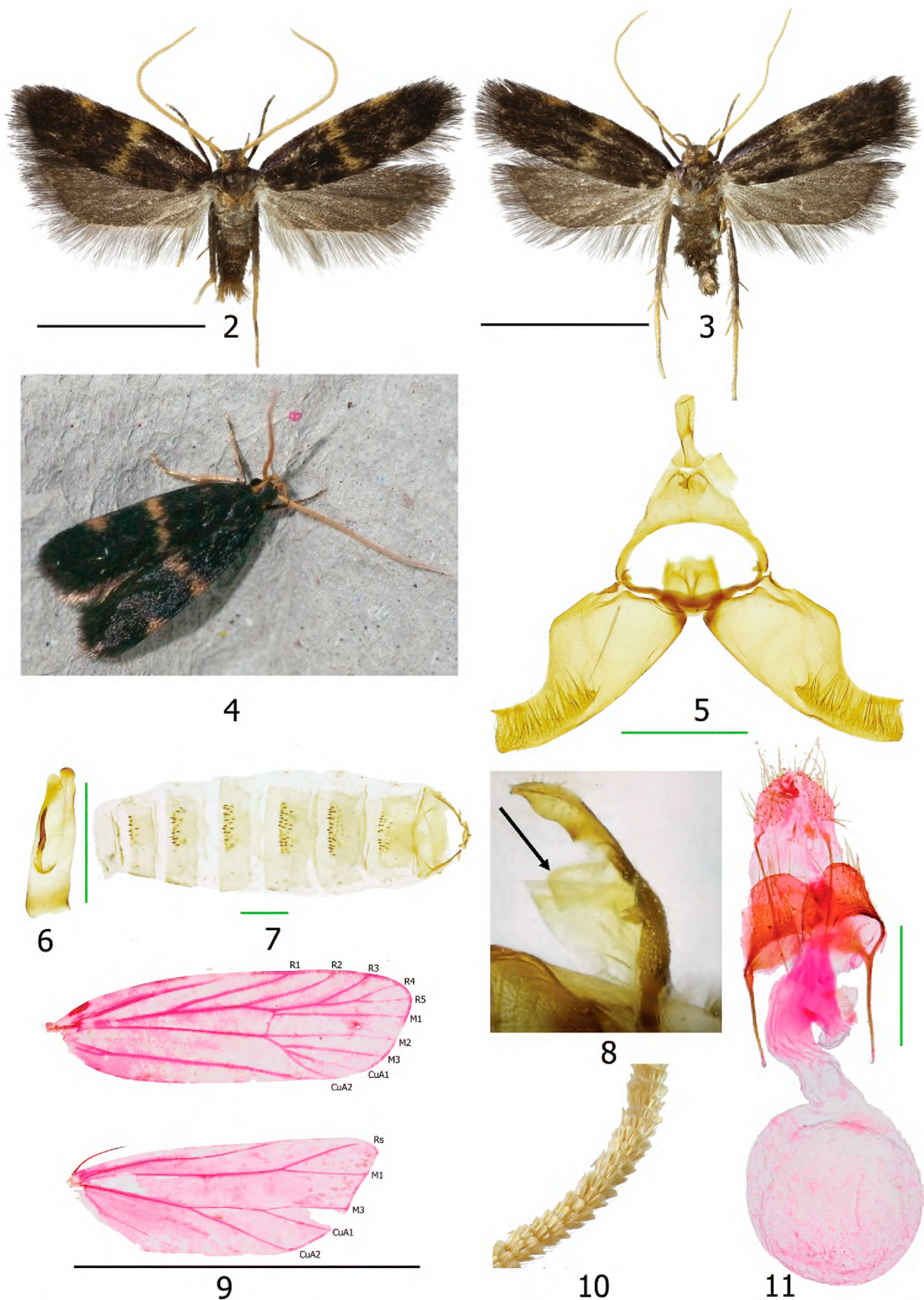
<https://zoobank.org/02DC9393-4978-4D54-B25C-29DFDE08B3E5>

Type material. *Holotype* ♂ Spain, Cádiz, Tarifa, CIMA, Site 1, MV light, 24.v.2022, 36.0167, -5.5864, 60 m, Leg. M. Sterling, D. Grundy, specimen no. NHMUK013698467, slide no. NHMUK014331156, wingspan 14 mm, fwl 6.25 mm. *Paratypes* (12♂, 3♀) 2♂, collection data as holotype, specimen number NHMUK013698468, specimen number NHMUK013698469; 1♀, 27.05.2022, otherwise same collection data for Site 1, specimen no. NHMUK013699868, slide no. NHMUK014331160. 3♂ Spain, Cádiz, Tarifa, CIMA, Site 2, MV light 25.v.2022, 36.0166, -5.5858, 75 m, specimen no. NHMUK013699866, slide no. NHMUK014331157, specimen no. NHMUK013699867, slide no. NHMUK014331158, specimen no. NHMUK013698979, slide no. NHMUK014331159; 1♀ 27.v.2022, otherwise same collection data for Site 2, specimen no. NHMUK013698523, slide no. NHMUK014331162. 3♂, Spain, Cádiz, Tarifa, CIMA, Site 3, MV light 27.05.2022 36.0148, -5.5871, 85 m, specimen no. NHMUK013699869, specimen no. NHMUK013699871, specimen no. NHMUK013698522; 1♀, same collection data as site 3, specimen no. NHMUK013699870. 3♂ Spain, Cádiz, Tarifa, CIMA, Site 4, MV light, 29.v.2022, specimen no. NHMUK013698524, specimen no. NHMUK013698525, specimen no. NHMUK013698526, 1♂ Site 4, 30.v.2022, specimen number NHMUK013699872. The holotype and 12 paratypes have been deposited at the NHMUK. In accordance with the terms of the collecting permit, the three last mentioned paratypes have been deposited with the Museo Nacional de Ciencias Naturales, Madrid.

Diagnosis. Forewings black with a purplish sheen, a narrow yellow bar from costa to dorsum before $\frac{1}{2}$ and a yellow costal spot at $\frac{3}{4}$ (Figs 2–4), antenna and labial palps yellow, antenna of male thickened with rings of large yellow scales projecting outwards from base of each flagellomere (Fig. 10). R3, R4 and R5 of forewing with a common stalk. M2 present in forewing but absent in hindwing. M3, CuA1 and CuA2 of forewing with a common stalk, CuA1 and CuA2 stalked in forewing (Fig. 9). Male genitalia with small membranous gnathos and very small sclerotised mesial process (Figs 5, 8), vinculum semicircular, sclerotised and melanised (Fig. 5). Female genitalia with appendix bursae, corpus bursae small and rounded without signum (Fig. 11).

Description. Male. Wingspan 13–15 mm. **Head.** Ocellus absent, frons dark grey, vertex dark grey with thick lateral tufts of dark grey and dark yellow scales, yellow scaling continuing above the eye; maxillary palps very small, grey, pilifers with thick brushes of short yellowish bristles. Labial palps long, thin, strongly recurved, approximately $3\times$ diameter of eye, dark yellow, basal segment paler than other segments, second and third segment same length. Antenna same length as forewing, yellow, long scape with appressed scales, rings of thick yellow scales pointing outwards from base of each flagellomere give antenna a thickened appearance (see Fig. 10). Haustellum with basal half scaled whitish. **Thorax.** Thorax and tegulae black. Foreleg dorsally dark grey, ventrally yellow, small tibial epiphysis, middle leg yellow with fine dark grey scales, tarsus of hind leg with fine dark grey scales, tibia and femur yellow with some darker scaling. **Wings.** Venation: Forewing with R3 stalked with stalk of R4 and R5, R4 and R5 stalked, M1 present, M2 and M3 almost parallel, M3 stalked with stalk of CuA1 and CuA2, CuA1 and CuA2 stalked. Hindwing with M2 absent (Fig. 9). Forewing costa slightly arched, apex rounded, termen angled inwards, tornus obtusely angled; ground colour black with a purplish sheen, thin yellow bar before $\frac{1}{2}$ from costa to dorsum, yellow patch on costa at $\frac{3}{4}$, cilia long, black. Hindwing with apex slightly projecting, dark grey, unmarked, cilia long, concolorous.

Female. Similar (including labial palps). Rings of antennal scales shorter and colour of antenna paler than in male.



Figures 2–11. *Xenotorodor stygioxanthus* gen. nov., sp. nov. **2.** ♂ Holotype. **3.** ♀ Paratype. **4.** ♂ Habitus. **5.** Male genitalia. **6.** Aedeagus. **7.** Pre-genital abdomen (**5–7.** ♂ holotype slide no. NHMUK014331156). **8.** ♂ Holotype lateral image prior to mounting, gnathos indicated. **9.** Wing venation slide no. NHMUK014331163. **10.** ♂ Holotype, highly magnified section of antenna. **11.** ♀ Paratype, female genitalia, slide no. NHMUK014331160. Scale bars: 5 mm (black); 0.5 mm (green). Figs **4**, **8** and **10** not to scale.

Male genitalia. Uncus directed caudally, narrow, elongate, slightly spatulate posteriorly, with narrow elongate lateral flanges. Gnathos fused to tegumen, small, box like, membranous, a very small sclerotised projection from tegumen medially. Tegumen broad anteriorly, tapering and indented posteriorly, weakly sclerotised, rough textured; pedunculi short, slightly curved. Vinculum semicircular, thin, strongly sclerotised, melanised. Saccus short and very broad. Juxta short and broad, almost square in shape, without caudal projections. Valva weakly sclerotised, costal margin bulging medially, sharply converging towards ventral margin postmedially, cucullus rectangular, small pointed projection at apex, cucullar region with dense broad setae, small digitate process near inner margin of setae. Aedeagus short, straight, weakly sclerotised, with a small rounded projection posteriorly, and a single broad arrow headed cornutus medially, strongly sclerotised posteriorly, hooked anteriorly.

Female genitalia. Papillae anales short and broad. Apophyses posteriores over $1.5\times$ length of apophyses anteriores. Eighth tergite rectangular. Eighth sternite indented posteriorly. Ostium circular. Antrum short, broad, membranous. Ductus bursae short, broad and membranous, appendix bursae present. Corpus bursae rounded, without signum.

Biology, behaviour and early stages. Collection of samples of leaf litter for early stages in May 2022 proved unsuccessful and the early stages are unknown. Since its discovery in 2020, the adult has been recorded in May, June, August, September and November. The principal emergences are in late May/June and August with 20 records between 18 May and 21 June 2020, 48 records between 24 May and 11 June 2022 and 22 records between 17 and 22 August 2021. Recording in May/June 2021 was not possible due to COVID restrictions. Both sexes are attracted to mercury vapour light and have been found flying around MV lights in the two hours after darkness. When the adult is resting the antennae are projected upwards and sideways (see Fig. 4) in an alert posture and tucked under the costa of the forewing when fully resting. The wings are always posteriorly flat to the resting surface (Fig. 4).

Distribution. The taxon is known principally from various localities in an area consisting of a patchwork of Mediterranean scrub, dry cattle grazed pasture, wild olive groves and some *Eucalyptus* sp. plantations, approximately 0.5 kilometres from the coast around the research centre at CIMA near the town of Tarifa in Southern Spain. Two specimens (not retained) were found in June 2022 at Huerta Grande, Pelayo (36.081, -5.503, 250 m) approximately 10 kilometres from CIMA and slightly further inland. Huerta Grande has been well recorded by DG in the flight period of this species for the last 10 years and the moth has never been recorded there before, so this is believed to be a new arrival. The climate in the presently known range is wet and warm in winter (temperatures not usually below freezing) and almost completely dry in summer.

Etymology. *Xenotorodor* from *xenos*, gr., meaning, among other things, stranger or outsider. This is a reference to the unusual combination of morphological features for a species of Torodorinae in the new taxon, and the substantial geographical extension of the range of the subfamily. The gender of the genus name is male. The specific name *stygiOXanthus* is from *stygios*, gr., meaning among other things extremely dark; and *xanthos*, gr., meaning yellow, a reference to the blackish forewings marked with yellow.

Material examined. Type material and six unset and unpinned specimens in tubes with the following data, which are excluded from the type material: 4♂ Spain, Cádiz, Tarifa, CIMA, MV light, 17–21.viii.2021, leg. D. Grundy; 2♂ Spain, Cádiz, Tarifa, CIMA, MV light, 05–13.xi.2021, leg. D. Grundy. The remainder of the specimens recorded were not retained although photographs of 20 of these specimens, taken by DG, were examined.

Discussion

Family placement

Park *et al.* (2022: 12, 14) note the following [syn-]apomorphies for Lecithoceridae: gnathos fused to tegumen, antennae usually longer than forewing and mesial process of gnathos of the male genitalia always downturned and laterally compressed. In the new taxon the gnathos is fused to the tegumen, the antennae are the same length as the forewing (Park *et al.* (2022: 15) note that a recent study of the Afrotropical fauna has confirmed that the antenna is not always longer than the forewing in several genera, including *Protolychnis*) but the gnathos of the new taxon is much reduced (Figs 5, 8). However, the subfamily Crocanthinae is defined as having the autapomorphic character with the gnathos always absent or reduced in the male genitalia (Park 2015: 252; Park *et al.* 2022: 16). At a family level, the reduced gnathos in the new taxon is therefore not inconsistent with a diagnosis of Lecithoceridae. The wing venation, including the stalking of R3, R4 and R5 and the stalking of CuA1 and CuA2 in the forewing and the absence of M2 in the hindwing is typical of a number of lecithocerid genera, as is the presence of tergal spines in the pre-genital abdomen. In the male genitalia, the narrow thorn like uncus, thin strongly sclerotised vinculum and the vestigial saccus are also often found in Lecithoceridae. The rings of outwardly pointing yellow scales arising from each flagellomere on the male antenna are also potentially synapomorphic with *Protolychnis* and *Sisyrodonta*, if those two taxa are found to belong to a clade not including *Halolaguna* (but see Fig. 1b).

The results from the DNA barcode query and the analyses considering distance and Neighbor Joining support a diagnosis of Lecithoceridae for the new taxon. Also, from our search of the sequences for synapomorphies, we consider the state Asparagine (N) in the 161st complete codon of the DNA barcode to represent a ground plan synapomorphy of Lecithoceridae, which is reversed in a few genera and species (for example distally in the genus *Sisyrodonta* and in *Lecithocera nigrana* and its widely dispersed species complex). The new taxon exhibits this synapomorphy. This is a groundplan feature of all lineages of Lecithoceridae *sensu* Wang and Li (2020), apart from their outgroup, *Martyringa* (USA-SE Asia), which exhibits either a Phenylalanine (F) or Serine (S) for this position, whereas *Ceuthomadarus*, *Synersaga* and *Nosphistica* all show an Asparagine in the homologous position.

Subfamily placement

According to Park (2022) the subfamily Ceuthomadarinae can be distinguished from the other subfamilies based on the absence of a proboscis and the subfamily Lecithocerinae can be defined and distinguished from Torodorinae by the presence of a bridge-like structure connecting the tegumen and the valval costa of the male genitalia. The new taxon has a well developed proboscis and does not have the bridge-like structure in the male genitalia used to distinguish Lecithocerinae from Torodorinae. The new taxon is therefore the first record of a new subfamily of Lecithoceridae for Europe. However, placement, on morphology, between the current concepts of Crocanthinae and Torodorinae is not as straightforward. On the basis of Park's definition of Crocanthinae, it would appear that the new taxon should be placed in Crocanthinae because it has a reduced gnathos. However, apart from the reduced gnathos, there is little in the adult morphology to connect the new taxon to Crocanthinae.

The suggested synapomorphies of Crocanthinae, apart from the state of the gnathos, include relatively bright coloured wings and hindwing often with similar markings like those of the forewing

(Park et al. 2022: 17). The new taxon has neither wing synapomorphy. Park (2015) redefines *Crocantthes* (the type genus of Crocanthinae) with the following morphological characters: labial palpus with dimorphism, male with second segment long, thickened with rough scales, and third segment absent, aborted or shortened, but female with normal slender third segment. Forewing normally elongate, with usually well-developed postmedian fascia; costa gently curved beyond 2/3; apex acute or normally produced; termen usually concave medially or slightly convex; venation with R2 usually free or sometimes short-stalked with R3 and R4; R5 absent; M2 absent; CuA1 and CuA2 short-stalked; cell opened. Hindwing usually unicolorous, as wide as forewing, apex acute; termen slightly concave or strongly oblique; venation with M2 absent; M3 and CuA1 stalked basally; CuA2 arising from the $\frac{1}{2}$ length of the wing. Apart from the configuration of the hindwing venation, the new taxon displays few of these characters. The labial palps are not dimorphic, in the male the second segment is not thickened with rough scales and the third segment is not absent, aborted or shortened, the forewings are not elongate and do not have a postmedian fascia and the apex is rounded. In the forewing venation, R3 is on a common stalk with R4 and R5, R5 is present and stalked with R4. M2 is present, the stalk of CuA1 and CuA2 is substantial and the cell is closed. In addition, the antennae in *Crocantthes* are not (with the exception of *C. diula*) thickened with outwardly projecting rings of scales. In the male genitalia of *Crocantthes*, the vinculum is U shaped, the juxta usually has caudal projections, the sacculus is developed and the cornuti in the aedeagus are complex.

In addition, the general appearance (externally and in the male and female genitalia) of the new taxon is substantially different from any species currently described within Crocanthinae, which are almost exclusively only known from the island of New Guinea and Australia, although a few genera reach other parts of Indonesia and the Philippines.

Only three existing genera of Crocanthinae are represented here (Fig. 1b) of which *Aprosoesta* tended to be the earliest diverging, and *Crocantthes* is also represented by its type species. The new taxon fell away from the sampled members of Crocanthinae when taxon sampling of identified terminals from BOLD was enhanced (as in Fig. 1b).

The subfamily Torodorinae is defined (Park et al. 2022) by the absence of a bridge-like structure connecting the tegumen and the valva and by the uncus usually thorn-like, directed caudally in the male genitalia. The present taxon has both these synapomorphies (although these are also present in most Crocanthinae). A comparison of the features of the new taxon with Park et al. (2022) shows that, although it has an unusual combination of features, it is the case that taxa with similarity to one or more characters among palps, antennae, wingshape, wing pattern, forewing venation and male genitalia, can be found within Torodorinae. In our view it is not inconsistent for a lecithocerid moth with a reduced gnathos to fall within Torodorinae. We also note that Yu et al. (2022) have recently noted 12 species of *Torodora* (the type genus of the subfamily) in which the gnathos is without a mesial process.

Moving to the molecular data, in interpreting the IQ-TREE analysis on the 19-terminal dataset (Fig. 1a) and based on the Phym1 (Fig. 1b) and MrBayes analyses (Suppl. materials 2, 3) of the full datasets, parts of the trees with good coverage of nuclear data (Lecithocerinae in particular with 10 taxa) show relatively good resolution, whereas other parts predominated by COI-5P (in which only *Thubana* cf. *deltaspis*, *Torodora aenoptera* and *Crocantthes prasinopis* included nuclear data) show relatively weak phylogenetic signal.

The IQ-TREE analysis (Fig. 1a) provides support for placement of the new taxon within a combined clade of Torodorinae + Crocanthinae (1/99), and no support for its placement in Lecithocerinae

or Ceuthomadarinae nor for a clade of *Xenotorodor* plus Crocanthinae (the last grouping occurred in the unlinked analysis, but without support). The analyses of the full dataset (Fig. 1b, Suppl. materials 2, 3) similarly provide no support for placement of the new taxon in Lecithocerinae or Ceuthomadarinae and, although the supporting data is almost all COI-5P, provide posterior support for the placement of the new taxon in a subclade of Torodorinae rather than Crocanthinae.

However, as noted in Park (2022), the subfamilial relationship of Lecithoceridae still reveals some problems. We confirm monophyly of Ceuthomadarinae (although inclusion of *Synersaga* or even *Nosphistica* is equivocal) and Lecithocerinae, although the relationship of these subfamilies varied among analyses and were sensitive to parameterisation, but linked analyses produced relationships consistent with that shown by Wang and Li (2020), with Lecithocerinae sister to Torodorinae + Crocanthinae. However, further work needs to be done to demonstrate that Torodorinae really constitutes a monophylum without the inclusion of all or part of Crocanthinae (i.e., whether proximal, distal, or sister to Crocanthinae in trees). Both the 17- and 19-terminal edge-unlinked IQ-TREE analysis showed support for a Crocanthinae + Torodorinae clade but the Torodorinae-only clade was unsupported by bootstrap in either analysis (or in Fig. 1b).

Generic placement

The morphological differences between *Xenotorodor* and *Crocantthes* are dealt with above. Also, the new taxon is morphologically divergent from all existing genera within Torodorinae and is genetically divergent from those eight traditional torodorine genera (plus *Sisyrodon*) whose DNA barcode sequences were available for analysis. It displays some distinctive characters shared with some genera (including *Sisyrodon* and *Cophomantella* which are not currently placed in either Torodorinae or Crocanthinae), but to place it in an existing genus would involve polyphyletic expansion of the concept of that genus. The genera with which the present taxon displays some potential synapomorphies (and the reason for rejecting placement in those genera) are:

1. *Protolychnis* Meyrick, 1925 (type species *Lecithocera maculata* Walsingham, 1881, from South Africa). The antenna of the type species was described by Lord Walsingham (Walsingham 1881) as thick yellowish ochreous. This character is present in most if not all species within the genus. The palps of the members of this genus are yellow and the antenna shorter than the forewing (Park *et al.* 2022: 189). The forewing pattern of *P. trigonias* (Meyrick, 1904) and *P. chlorotoma* (Meyrick, 1914) have some similarities to the present taxon (although the hindwings are pale in *P. trigonias*) and the forewing and hindwing venation are similar. However, in the male genitalia, the gnathos is developed with a large mesial process, and in the type species and most other species the saccus is developed, the juxta has caudal projections, the aedeagus is slender, usually longer than the valva and the female genitalia has a horseshoe shaped signum (Park *et al.* 2022: 190). The female genitalia also lack an appendix bursae. Further, although the venation is similar, M2 and M3 are approximated in the forewing in *Protolychnis* whereas in the present taxa they are almost parallel (see Park and De Prins (2019), Park and Koo (2020, 2022) and Park *et al.* (2022) for further information). DNA barcodes of *Protolychnis* show a phylogenetically close, but not sister relationship with the new taxon (Fig. 1b). A DNA barcode has been identified as the type species (*P. maculata*; BOLD:AAK9877; HM860216.1), and there are at least two closely related taxa widespread in Africa on BOLD (BOLD:ADT8222, not shown, is 4.1% pairwise divergent from the *P. maculata* sequence). Nevertheless, the sequence of the new taxon OQ339151 (NHMUK013698467) exhibits a 11.4% pairwise divergence to HM860216.1. This seems a consid-

erable distance for a potential congeneric. It is greater than for the highly allopatric *P. lychnocentra* from Australia (which is clearly a *Protolychnis* according to Park et al. 2022) and which differs from *P. maculata* by 10.3% and *X. stygiexanthus* by 11.52%, and a likely member of the genus from China (BOLD:ACD5139; Fig. 1b) which differs from *P. lychnocentra* by 6.5% and *P. maculata* by 9.6% (Fig. 1b). *P. trigonias* (Meyrick, 1904) (BOLD:AAO4343; also Australia) did not group with the two other identified *Protolychnis* in trees; this taxon is 12.64% pairwise divergent from *X. stygiexanthus*; 610 bp) and 13.1% pairwise divergent from *P. maculata*; rather, it groups with *Athymoris martialis* Meyrick, 1935 and a species of “*Cophomantella*” (BOLD:AAL6994) (Fig. 1b, Suppl. material 2). It therefore seems doubtful that *P. trigonias* represents a true *Protolychnis*, although the missing abdomen of the type (*Styloceros trigonias* Meyrick, 1904) has made morphological assessment difficult (Park et al. 2022: 196).

2. *Sisyrodonta* Meyrick, 1922 (type species *Sisyrodonta ochrosidera* Meyrick, 1922, from Australia). This is currently a monotypic genus known from Western Australia, although DNA barcodes indicate (e.g. Fig. 1b) that there is a species complex widely spread through Australia (BOLD:AAJ6848, BOLD:ACK2022; identified as a *Crocantthes*, BOLD:AAN9227, BOLD:AAO6600). The forewing pattern of *S. ochrosidera* is black (with a purplish sheen) and yellow, the antenna and labial palps are yellow and each flagellomere of the male antenna is ringed with large yellow scales. The antenna is more lamellate than in the present taxon and Meyrick considered it to be unique (Meyrick 1922) but in our view the antennae are similar to those of the new taxon. However, the hindwings are pale in both the type species and undescribed members of the complex. More importantly, CuA1 is absent in the forewing in the genus and CuA2 and M3 are stalked in the type species. In the male genitalia, the uncus of *S. ochrosidera* is broad at base and strongly sclerotised throughout, the gnathos has a large, strongly sclerotised medial section which is strongly beaked and strongly projected posteriorly and in the specimen of *Sisyrodonta* sp. from Western Australia (NHMUK013698527; Slide no. NHMUK014331161) which we have examined, the aedeagus is thin and elongate and without a cornutus. *Sisyrodonta* was included in the ‘Gelechiidae’ by Fletcher (1929: 204). It was transferred to the Lecithoceridae by Sattler (1973: 250). The genus was transferred to the Xyloryctidae from the Lecithoceridae by Gozmány (1978: 263). However, Common (1990: 264) considered that the wing venation, the presence of the supplementary wing-coupling setae, and the behaviour of the adults leaves little doubt that it belongs in the Lecithoceridae. *Sisyrodonta* was also treated in the Lecithoceridae by Nye and Fletcher (1991: 277). We consider that *Sisyrodonta* is correctly placed in the Lecithoceridae although the strongly sclerotised medial section of the gnathos is unusual for the family. We place the genus, which in the full dataset analyses (see Fig. 1b, Suppl. material 3) fell in a small grouping that included *Protolychnis* and *Halolaguna*, as well as the new taxon, in the Torodorinae.

3. *Cophomantella* Fletcher, 1940 (type species *Onebala elaphopis* Meyrick, 1910, from India). *Cophomantella* was established as an objective replacement name for *Cophomantis*, Meyrick, 1925, a junior homonym of a frog, *Cophomantis* Peters, 1870 (Nye and Fletcher 1991). As a result of various combinations, “*Cophomantella*” applies to a few remaining species and Park et al. (2022: 24) recommend that the type species should be transferred to a genus in Gelechiidae. We have examined the type of *C. elaphopis*, including the wing preparation and male genitalia contained on the type slide (JFGC 8911), and apart from the reduced gnathos and somewhat reduced venation, it exhibits many of the typical features of a torodorine including the tergal spines on the abdomen, the thorn-like uncus directed caudally, the shape and orientation of the valva and the large, complex cornuti in the aedeagus. We therefore consider *Cophomantella* to be a further

example of a genus within Torodorinae with a reduced gnathos, and we transfer it back to Lecithoceridae. The new taxon is not, however, within *Cophomantella*. The palps of *C. elaphopis* are dark, the scaling on the antennae of the male is not projected outwards, the forewing markings are different and in the forewing venation, R3 and R4 are stalked, R5 is absent and M3 is absent. Also, in the male of *C. elaphopis* there is an expansible pencil of hairs from the base on the forewing ventral surface along the costa (Meyrick 1910). This is not present in the new taxon. In the male genitalia of *C. elaphopis*, the vinculum is U shaped and the tegumen and valvae are also a different shape from those of the new taxon. The one South East Asian BIN on BOLD attributed to the genus (BOLD:AAL6994) that we analysed fell with posterior support together with *Protolychnis trigonias* from Australia (Fig. 1b; but see also Suppl. material 3).

4. *Torodora* Meyrick, 1894 (type species *Torodora characteris* Meyrick, 1894, from Myanmar). This is a genus which comprises more than 200 described species (Park *et al.* 2022: 271) and seems very likely to be polyphyletic. It is considered here on the basis of the resemblance of several features in the male genitalia, including the shape of the uncus, juxta, vinculum, saccus and valva and the rough texture of the tegumen in *T. meifengensis* Park, 2015; *T. octavana* (Meyrick, 1911); and *T. umbriella* Park & Heppner, 2022 (Fig. 5, Park *et al.* 2022 plates 113C, 118E, 132A). The genus is generally defined by the wing venation as follows: Forewing with R3, R4 and R5 usually on a common stalk, CuA1 and CuA2 stalked, and M2 present in both wings (Park *et al.* 2022, 271). The present taxon satisfies these criteria except that M2 is absent in the hindwing, although Park *et al.* (2022: 272) notes that venation is variable and that generic assignment should be based on a combination of characters. According to Park, the male genitalia of *Torodora* are characterised by the hooked gnathos, the foot shaped or variously elongated valva and the strong spinous zones on the tergites of the abdomen (Park *et al.* 2022: 272), although Yu *et al.* (2022) give mesial process of the gnathos as present or absent as a generic character and identify 12 species of *Torodora* in which it is absent. The latter two characters are present in the new taxon (the second occurs in the ground plan of all Lecithoceridae, Autostichidae and Xyloryctidae) but the new taxon lacks the hooked mesial process of the gnathos (which is present in the three species of *Torodora* referred to above). In addition, the wing pattern and thickened antenna in the male would be unusual for *Torodora*. The one identified species we analysed, *T. aenoptera*, fell in an analysis (Fig. 1b) within Torodorinae, next to a species from Australia (BOLD:AAH3804), and a species of *Parkiana* from Madagascar (see Park *et al.* 2020), however, with no support for its placement.

5. *Thubana* (Type species *Thubana bisignatella* Walker, 1864, from Borneo). Although it is 9.2–9.38% pairwise divergent, *T. exoema* (Meyrick, 1911) is the nearest identified sequenced species of Lecithoceridae to the present taxon in the NJ tree resulting from the BOLD search, while the one *Thubana* species analysed (Figs 1a, 1b) also shows no sister relationship to it. According to Park *et al.* (2022: 212), the genus is characterised by the following synapomorphic characters: the forewing commonly having a large creamy-white or light-orange triangular costal patch, with a few exceptions, and the venation with M3, CuA1, and CuA2 on a common stalk; R3 stalked with R4 and R5; R5 absent or often present; the hindwing with M2 present and closely approximated to the stalk of M3 and CuA1; the abdomen with dense spinous zones on tergites. The present taxon does not have the triangular costal patch, R5 is present in the forewing and M2 is absent in the hindwing. The tergal spines are moderate rather than dense. The male genitalia of *Thubana* are significantly different. The gnathos is strongly developed with a large downward mesial projection, the vinculum is U shaped and projects well beyond the base of the valva, the juxta has caudal projections and

the cornuti in the aedeagus are complex and not a single spike. In the female genitalia the ductus bursae is long and usually with sclerotisation or scobination and the corpus bursae has a signum.

6. *Halolaguna* (Type species *Halolaguna subluxata* Gozmany, 1978, from China). This is one of the genera which appears in a molecular clade of Torodorinae with the present taxon (along with typical *Protolychnis* and *Sisyrodonta*, Fig. 1b, pp = 0.99, albeit without support for its sister taxon, and in a clade that has posterior support (pp = 0.97) in the linked rather than unlinked MrBayes analysis, see Suppl. material 3). Morphologically there is little similarity with this genus. In *Halolaguna* the apex of the forewing projects and the wing pattern, antennae and colour of the labial palps are different. M2 is also present in the hindwing. In the male genitalia the gnathos is developed with a strong downward mesial projection, the vinculum projects beyond the base of the valva, the saccus is developed, the juxta has caudal projections and the cornuti in the aedeagus are complex with minute spines. In the female genitalia the ductus bursae is long and the corpus bursae has a signum.

7. *Crocantthes* (Type species *Crocantthes prasinopis* Meyrick, 1886: 277, from Australasia) and *Aprosoesta* Turner, 1919 (type species *A. pancala* Turner, 1919). See above for discussion of *Crocantthes*. *Aprosoesta* is mentioned separately but only because of the relative proximity of the *A. pancala* sequences to those of the new taxon in Fig. 1b. *Aprosoesta* was previously treated as a junior synonym of *Crocantthes* but was re-established in Park 2015 on the basis of the similar maculations of the forewing and hindwing, and characters in the forewing venation and the third segment of the labial palps in the male. In terms of morphology, *Aprosoesta* is as divergent as *Crocantthes* from the new taxon (*A. 'pancala'* BOLD: AAO6447 is about 11.5% divergent by nucleotides).

The origin of the population of this taxon in Southern Spain is not clear. It could be a previously undiscovered but long resident population. Although the Microlepidoptera of Spain seem reasonably well studied, there must be many species still awaiting discovery. However, this is a distinctive taxon, evidently common where it occurs, which is not likely to be overlooked and the habitat in which it occurs is not uncommon in Southern Spain so there is no obvious reason why it would not occur elsewhere. A second possibility is that it is a previously unknown species originating from North Africa which is now spreading into Southern Europe (although it is clearly distinct morphologically and genetically from *Ceuthomadarus* from NW Africa). A number of species have been found around Tarifa which are taking this route, most likely as a result of climate change making conditions hotter and harsher in North Africa, causing resident species to expand northwards. DG has recorded in this area since 2017 but did not see this taxon until May 2020. Considering the increasing numbers of *X. stygioxanthus* at the locations around CIMA and the discovery of specimens at Huerta Grande, arrival from Africa may explain the Spanish population. A third possibility is that this taxon is an invasive species that has come, possibly from Australia, as a result of plantation of *Eucalyptus* spp. or other imported plants in the area. However, the taxon does not seem to be closely related to any known or previously DNA barcoded Australian genus of lecithocerid moth, and the DNA barcoding campaign on ANIC collections was more extensive than for any comparable tropical region.

Conclusions

It is noteworthy to detect a new subfamily for the European continent that is established in Spain. This interesting taxon from around Tarifa exhibits considerable morphological and genetic

divergence from hitherto known or DNA barcoded taxa (between about 10 and 13% to its nearest phylogenetic neighbours, and more than 9.2% from nearest hits as regards sequences not compromised by ambiguity codes). Its placement was not straightforward. It is clearly a lecithocerid both on the basis of its morphology and its DNA barcode, and it appears to fall by phylogenetic analysis within Torodorinae rather than Crocanthinae, despite its reduced gnathos. Our placement in a new genus is by elimination and should spur other attempts to find its closest relatives. The work we have carried out in seeking to place the taxon shows that existing data is insufficient to show that Torodorinae and Crocanthinae are separate clades, with an expanded phylogenomic dataset clearly needed, and that the morphological synapomorphies supporting Lecithoceridae, Torodorinae and Crocanthinae need further refinement. Torodorinae is a subfamily which is widespread palaeotropically and we are unable to narrow the origin of *X. stygioxanthus*. Further field and taxonomic studies of this taxon and other Lepidoptera in adjacent parts of North Africa and Southern Spain are likely in our view to yield interesting results in terms of taxonomic diversity, relationships of the currently established fauna, and changes in populations as a result of changing climate conditions.

Mandatory statements

The specimens examined for this study were collected in accordance with a permit issued by the Junta de Andalucia dated 11 April 2022. No specific funding was provided for this project and there are no conflicts of interest.

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References

- Barton I (2015) A contribution to the microlepidopteran fauna of Cyprus. *Entomologist's Record and Journal of Variation* 127(4): 157–167.
- Common IBM (1990) *Moths of Australia*. Melbourne University Press, Melbourne, 535 pp.
- Cuber P, Chooneea D, Geeves C, Salatino S, Creedy TJ, Griffin C, Sivess L, Barnes I, Price B, Misra R (2023) Comparing the accuracy and efficiency of third generation DNA barcode sequencing: Oxford Nanopore Technologies versus Pacific Biosciences. SSRN. <http://dx.doi.org/10.2139/ssrn.4353430>

- Fletcher TB (1929) A list of generic names used for the Microlepidoptera. *Memoirs of the Department of Agriculture of India (Entomology)* 11: 1–244.
- Fletcher TB (1940) New generic names for Microlepidoptera. *Entomologist's Record and Journal of Variation* 52: 17–19.
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3(5): 294–299.
- Gozmány LA (1978) Lecithoceridae. In: Amsel HG, Gregor F, Reisser H (Eds) *Microlepidoptera Palaearctica*, 5. Verlag Georg Fromme and Co., Wien, Vienna, 1–306.
- Gozmány LA (2012) *Lepidoptera of Greece and Cyprus Vol. 1. Fauna Graeciae* 9: 1–409.
- Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* 41: 95–98.
- Kaila L, Mutanen M, Nyman T (2011) Phylogeny of the mega-diverse Gelechioidea (Lepidoptera): adaptations and determinants of success. *Molecular Phylogenetics and Evolution* 61(3): 801–809. <https://doi.org/10.1016/j.ympev.2011.08.016>
- Klots AB (1970) Lepidoptera. In: Tuxen SL (Ed.) *Taxonomist's Glossary of Genitalia in Insects*. Munksgaard, Copenhagen, 115–130.
- Kristensen NP (2003) Skeleton and muscles: adults. In: Kristensen NP (Ed.) *Lepidoptera, moths and butterflies. Vol. 2. Morphology, physiology, and development. Handbook of Zoology IV* (36). Walter de Gruyter, Berlin, New York, 39–131. <https://doi.org/10.1515/9783110893724.39>
- Meyrick E (1894) On a collection of Lepidoptera from Upper Burma. *Transactions of the Entomological Society of London* 1894(1): 1–29.
- Meyrick E (1904) Descriptions of Australian Micro-Lepidoptera. *Proceedings of the Linnean Society of New South Wales* 29(2): 255–440. <https://doi.org/10.5962/bhl.part.20163>
- Meyrick E (1910) Descriptions of Indian Microlepidoptera. *Journal of the Bombay Natural History Society* 20(2): 435–462.
- Meyrick E (1911) Descriptions of Indian Microlepidoptera. *Journal of the Bombay Natural History Society* 20(3): 706–736.
- Meyrick E (1922) Results of Dr. E Mjöberg's Swedish scientific expedition to Australia 1910–1913. 27. Microlepidoptera. *Arkiv for Zoologi* 14(15): 1–13. <https://doi.org/10.5962/bhl.part.20146>
- Nye IWB, Fletcher DS (1991) *The Generic Names of Moths of the World. Vol. 6. Microlepidoptera*. Natural History Museum (London) Publications, London, 368 pp. <https://doi.org/10.5962/bhl.title.119516>
- Park K-T (2015) A new subfamily Crocanthinae based on the genus *Crocantthes* Meyrick and its related genera, with a world catalog of the subfamily (Lepidoptera, Lecithoceridae). *Journal of Asia-Pacific Biodiversity* 8: 251–286. <https://doi.org/10.1016/j.japb.2015.10.010>
- Park K-T (2022) Global biodiversity of the family Lecithoceridae (Gelechioidea) with a brief historical review. *Diversity* 14: 822. <https://doi.org/10.3390/d14100822>
- Park K-T, Cho S, Koo J-M (2022) *The subfamily Torodorinae of the World (Lepidoptera: Lecithoceridae)*. National Institute of Biological Resources, Incheon, Republic of Korea, 584 pp.
- Park K-T, Heppner JB (2022) Notes on Taiwan moths, 10. New species of *Torodora* from Taiwan (Lepidoptera: Lecithoceridae: Torodorinae). *Lepidoptera novae* 15(1–2): 19–25.
- Park K-T, Heppner JB, Bae Y-S (2015) Two new species of *Torodora* Meyrick (Lepidoptera, Lecithoceridae) and two species new to Taiwan. *Proceedings of the Entomological Society of Washington* 117(2): 183–193. <https://doi.org/10.4289/0013-8797.117.2.183>
- Park K-T, Koo J-M, Minet J (2020) Review of the Malagasy lecithocerid species described by Pierre Viette and deposited in MNHN (Paris), with new generic combinations and descriptions of a new subfamily and genus of Momphidae (Lepidoptera: Gelechioidea). *Zootaxa* 4845(2): 151–190. <https://doi.org/10.11646/zootaxa.4845.2>

- Regier JC, Mitter C, Zwick A, Bazinet AL, Cummings MP, Kawahara A, Sohn J-C, Zwickl DJ, Cho S, Davis DR, Baixeras J, Brown J, Parr C, Weller S, Lees DC, Mitter K (2013) A large-scale, higher-level, molecular phylogenetic study of the insect order Lepidoptera (moths and butterflies). PLoS ONE 8(3): e5856. <https://doi.org/10.1371/journal.pone.0058568>
- Robinson GS (1976) The preparation of slides of Lepidoptera genitalia with special reference to the Microlepidoptera. Entomologist's Gazette 27: 127–132.
- Sattler K (1973) A catalogue of the family-group and genus-group names of the Gelechiidae, Holcopogonidae, Lecithoceridae and Symmocidae (Lepidoptera). Bulletin of the British Museum (Natural History) Entomology 28: 153–282.
- Srivathsan L, Katoh K, Hartop E, Kutty SN, Wong J, Yeo D, Meier R (2021) ONTbarcoder and MinION barcodes aid biodiversity discovery and identification by everyone, for everyone. BMC Biology 19: 217. <https://doi.org/10.1186/s12915-021-01141-x>
- Sterling MJ, Sattler K, Lees DC (2022) *Isophrictis anthemidella* (Wocke, 1871) (Lep.: Gelechiidae), new for Great Britain. Entomologist's Record and Journal of Variation 134(7): 7–15.
- Walsingham (1881) Lord Walsingham on the Tortricidae. Transactions of the Entomological Society of London 1881(2): 219–288. <https://doi.org/10.1111/j.1365-2311.1873.tb01522.x>
- Wang Q-Y, Li H-H (2020) Phylogeny of the superfamily Gelechioidea (Lepidoptera: Obectomera), with an exploratory application on geometric morphometrics. Zoologica Scripta 49: 307–328. <https://doi.org/10.1111/zsc.12407>
- Yu S, Zhu Y, Wang S (2022) Eighteen new species and fifteen new records of the genus *Torodora* Meyrick (Lepidoptera: Lecithoceridae) from China. Zootaxa 5133(1): 1–39. <https://doi.org/10.11646/zootaxa.5133.1.1>

Supplementary material 1

DNA sequences

Authors: David C. Lees

Data type: table (Excel spreadsheet)

Explanatory note: DNA sequences for terminals used in the trees (Fig. 1a, b; Suppl. materials 2, 3) with any changes in identification, BIN (or if not available distance to nearest BIN), Process Ids, GenBank Accession numbers for each of seven genes, prior publication/link to NCBI and/or permission to use.

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Link: <https://doi.org/10.3897/nl.46.101457.suppl1>

Supplementary material 2

50% majority rule consensus tree 1

Authors: David C. Lees

Data type: figure (tif image)

Explanatory note: 50% majority rule consensus tree for a partitioned analysis of the 51 taxa, 5408 bp dataset in MrBayes 3.1. Support values are posterior probabilities based on a subset of 7,500 trees out of 15,001 sampled every 1,000 generations per separate run, by which point the runs had converged to 1.000. All parameters (except branch length) were unlinked and a variable rate prior was applied across the seven partitions and the final log likelihood for the best state of each ‘cold’ run asymptoted at - 28054 or - 28059.

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Link: <https://doi.org/10.3897/nl.46.101457.suppl2>

Supplementary material 3

50% majority rule consensus tree 2

Authors: David C. Lees

Data type: figure (tif image)

Explanatory note: 50% majority rule consensus tree for a partitioned analysis of the 51 taxa, 5408 bp dataset in MrBayes 3.1. Support values are posterior probabilities based on a subset of 7,463 trees out of 15,000 sampled every 1,000 generations per separate run, by which point the runs had converged to 1.000. All parameters were linked (by default) and a variable rate prior was applied across the seven partitions and the final log likelihood for the best state of each ‘cold’ run asymptoted at -28634 or -28635.

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